

# Cacnb2 Cas9-KO Strategy

**Designer:** 

**Reviewer:** 

**Design Date:** 

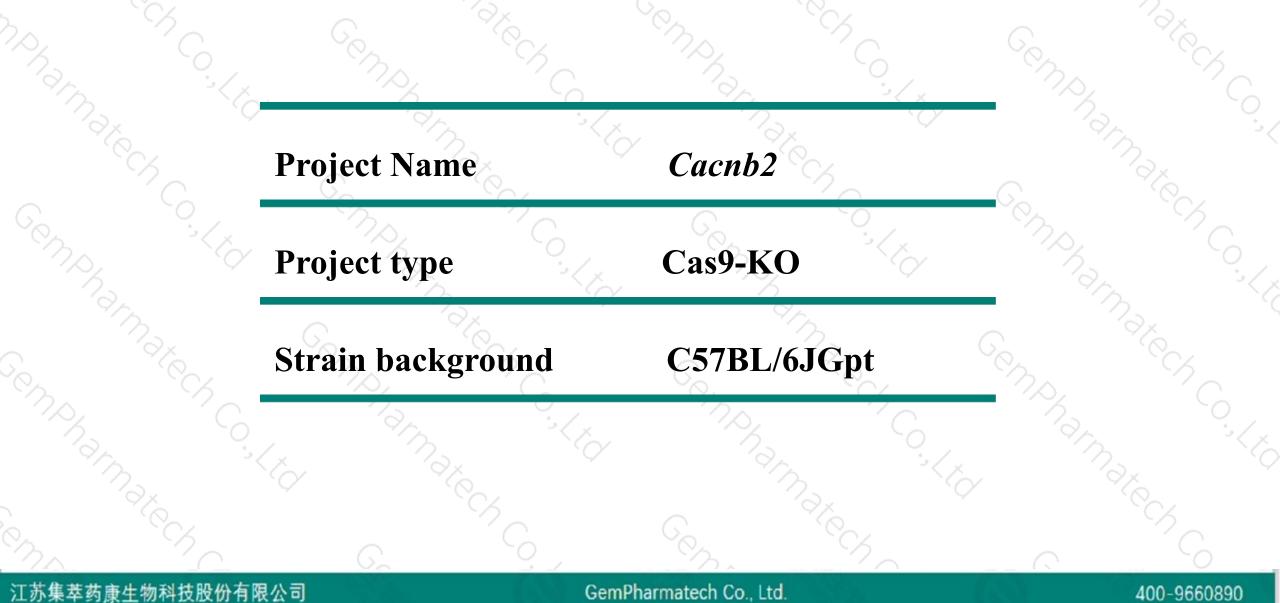
Daohua Xu

Huimin Su

2020-5-26

### **Project Overview**

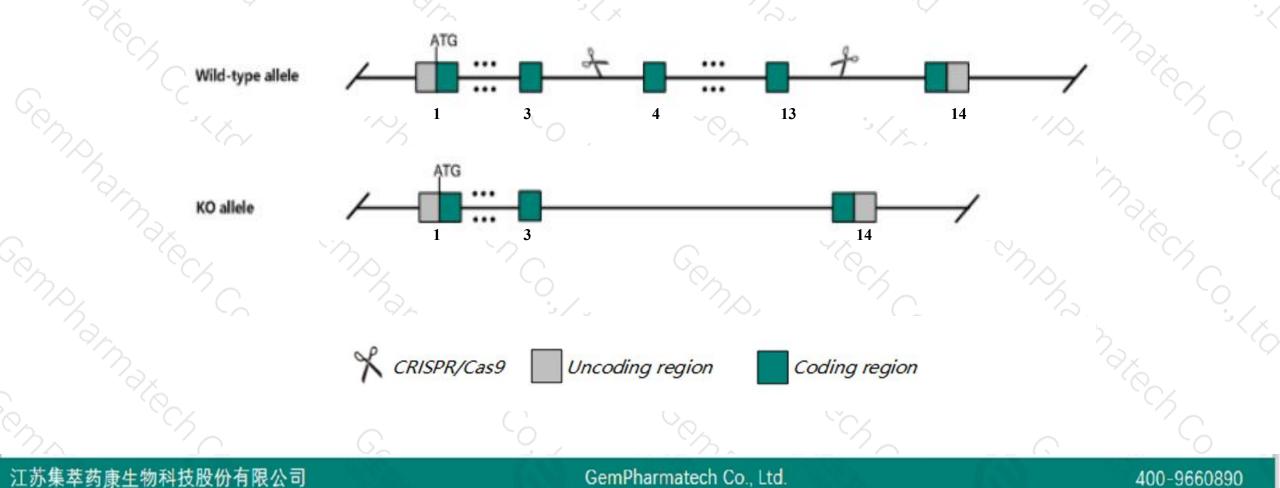




## **Knockout** strategy



This model will use CRISPR/Cas9 technology to edit the Cacnb2 gene. The schematic diagram is as follows:





- The Cacnb2 gene has 16 transcripts. According to the structure of Cacnb2 gene, exon4-exon13 of Cacnb2-203 (ENSMUST00000114723.8) transcript is recommended as the knockout region. The region contains 1158bp coding sequence. Knock out the region will result in disruption of protein function.
- > In this project we use CRISPR/Cas9 technology to modify Cacnb2 gene. The brief process is as follows: CRISPR/Cas9 system

- According to the existing MGI data, mice homozygous for a null allele exhibit lethality at e10.5 with growth retardation, abnormal yolk vasculature and abnormal cardiac development and function.
   The KO region contains functional region of the *Gm*37891 gene Knockout the region may affect the function of the *Gm*37891 gene Knockout the region may affect the function of the *Gm*37891 gene Knockout the region may affect the function of the *Gm*37891 gene Knockout the region may affect the function of the *Gm*37891 gene Knockout the region may affect the function of the *Gm*37891 gene Knockout the region may affect the function of the *Gm*37891 gene Knockout the region may affect the function of the *Gm*37891 gene Knockout the region may affect the function of the *Gm*37891 gene Knockout the region may affect the function of the *Gm*37891 gene Knockout the region may affect the function of the *Gm*37891 gene Knockout the region may affect the function of the *Gm*37891 gene Knockout the region may affect the function of the *Gm*37891 gene Knockout the region may affect the function of the *Gm*37891 gene Knockout the region may affect the function of the *Gm*37891 gene Knockout the region may affect the function of the *Gm*37891 gene Knockout the region may affect the function of the *Gm*37891 gene Knockout the region may affect the function of the *Gm*37891 gene Knockout the region may affect the function of the *Gm*37891 gene Knockout the region may affect the function of the *Gm*37891 gene Knockout the region may affect the function of the *Gm*37891 gene Knockout the region for the *Gm*37891 gene Knockout the region may affect the function of the *Gm*37891 gene Knockout the region for the *Gm*37891 gene Knockout the region for the *Gm*37891 gene Knockout the *Gm*37891
- The KO region contains functional region of the Gm37891 gene. Knockout the region may affect the function of Gm37891 gene.
- ≻The KO region deletes most of the coding sequence, but does not result in frameshift.
- The Cacnb2 gene is located on the Chr2. If the knockout mice are crossed with other mice strains to obtain double gene positive homozygous mouse offspring, please avoid the two genes on the same chromosome.
- This strategy is designed based on genetic information in existing databases.Due to the complexity of biological processes, all risk of the gene knockout on gene transcription, RNA splicing and protein translation cannot be predicted at the existing technology level.

Notice

### **Gene information** (NCBI)



☆ ?

Cacnb2 calcium channel, voltage-dependent, beta 2 subunit [Mus musculus (house mouse)]

Gene ID: 12296, updated on 20-Mar-2020

#### - Summary

 Official Symbol
 Cacnb2 provided by MGI

 Official Full Name
 calcium channel, voltage-dependent, beta 2 subunit provided byMGI

 Primary source
 MGI:MGI:894644

 See related
 Ensembl:ENSMUSG0000057914

 Gene type
 protein coding

 VALIDATED
 VALIDATED

 Organism
 Mus musculus

 Lineage
 Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha;<br/>Muroidea; Murinae; Mus; Mus

 Also known as
 AW060387, CAB2, Cavbeta2, Cchb2

 Expression
 Broad expression in heart adult (RPKM 6.4), cerebellum adult (RPKM 5.3) and 18 other tissues

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### **Transcript information (Ensembl)**



#### The gene has 16 transcripts, all transcripts are shown below:

p Protein Bio	type CCDS	UniProt	Flags
945 <u>655aa</u> Proteir	coding <u>CCDS15702</u>	Q32MF3 Q8CC27	TSL:1 GENCODE basic
117 <u>611aa</u> Proteir	coding <u>CCDS84473</u>	Q8CC27	TSL:1 GENCODE basic APPRIS is a system to annotate alternatively spliced transcripts based on a range of computational methods to identify the most functionally important transcript(s) of a gene. APPRIS ALT
117 <u>605aa</u> Proteir	coding <u>CCDS79735</u>	<u>C7IVS7</u>	TSL:1 GENCODE basic APPRIS is a system to annotate alternatively spliced transcripts based on a range of computational methods to identify the most functionally important transcript(s) of a gene. APPRIS PS
03 <u>72aa</u> Proteir	coding -	A0A0A6YY52	TSL:3 GENCODE basic
30 <u>143aa</u> Proteir	coding -	A0A0A6YWX5	5' and 3' truncations in transcript evidence prevent annotation of the start and the end of the CDS. CDS 5' and 3' incomplete TSL:5
146 No protein Processe	d transcript -	-	TSL:5
030 No protein Retain	ed intron -	-	TSL:2
457 No protein Retain	ed intron -	25	TSL:1
802 No protein Retaine	ed intron -		TSL:5
144 No protein Retaine	ed intron -	-	TSL:1
526 No protein Retain	d intron -	-	TSL:NA
052 No protein Retaine	d intron -	25	TSL:5
42 No protein Retain	ed intron -	-	TSL:5
35 No protein Retaine	d intron -		TSL:3
67 No protein Retain	ed intron -	-	TSL:5
69 No protein Retain	ed intron -	26	TSL:1
		-	

The strategy is based on the design of *Cacnb2-203* transcript, the transcription is shown below:

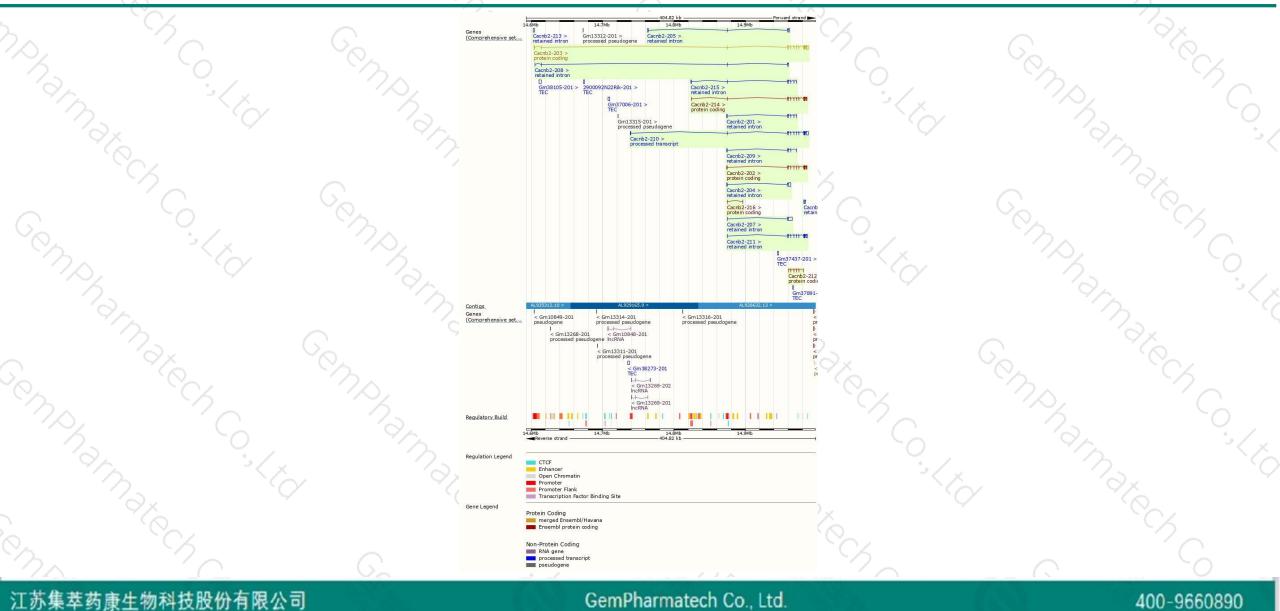


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### **Genomic location distribution**





### **Protein domain**

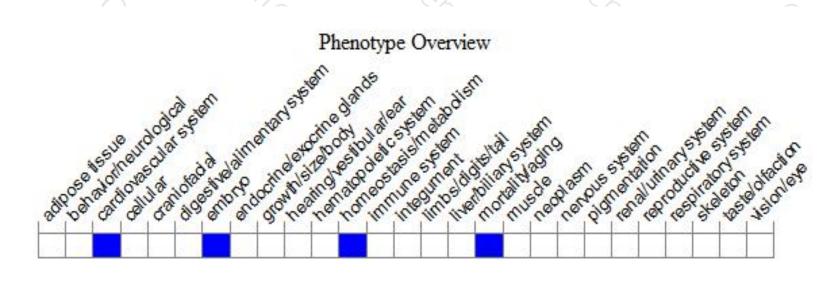
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5	Scale bar	0	60	120	180	240	300	360	420	480	540	655	0
	Variant Legend		nissense va ynonymou										;
	All sequence SNPs/i	Seque	since varian			sources)	- 1	1	1	1.1	1 11 11	1	10
	CDD	Conu	ance varian	CACNB2, s	SH3 domain								0
10	Gene3D	1	- 0.0 A P	),30,40		3,40,	50,300						X
	PANTHER		THR11824	S.F9									
	PROSITE profiles			SH3 doma	in	C	iuanylate kir	nase/L-type	calcium ch	annel beta	subunit		
	Pfam.		Volta	Voltage ge-dependent	1.5. State 1. State 1		, beta subur	nit					'S
	Prints				_		ltage-depen			L-type, be	ta subunit		
8	SMART						Suanylate kir	ase/L-type	calcium ch	annel beta	subunit		
9	Superfamily		SH3-like domain superfamily P-loop containing nucleoside triphosphate hydrolase										. /
	MobiDB lite Low complexity (Seg)									_			

### Mouse phenotype description(MGI)





Phenotypes affected by the gene are marked in blue. Data quoted from MGI database(http://www.informatics.jax.org/).

According to the existing MGI data, mice homozygous for a null allele exhibit lethality at E10.5 with growth retardation, abnormal yolk vasculature and abnormal cardiac development and function.



If you have any questions, you are welcome to inquire. Tel: 400-9660890



